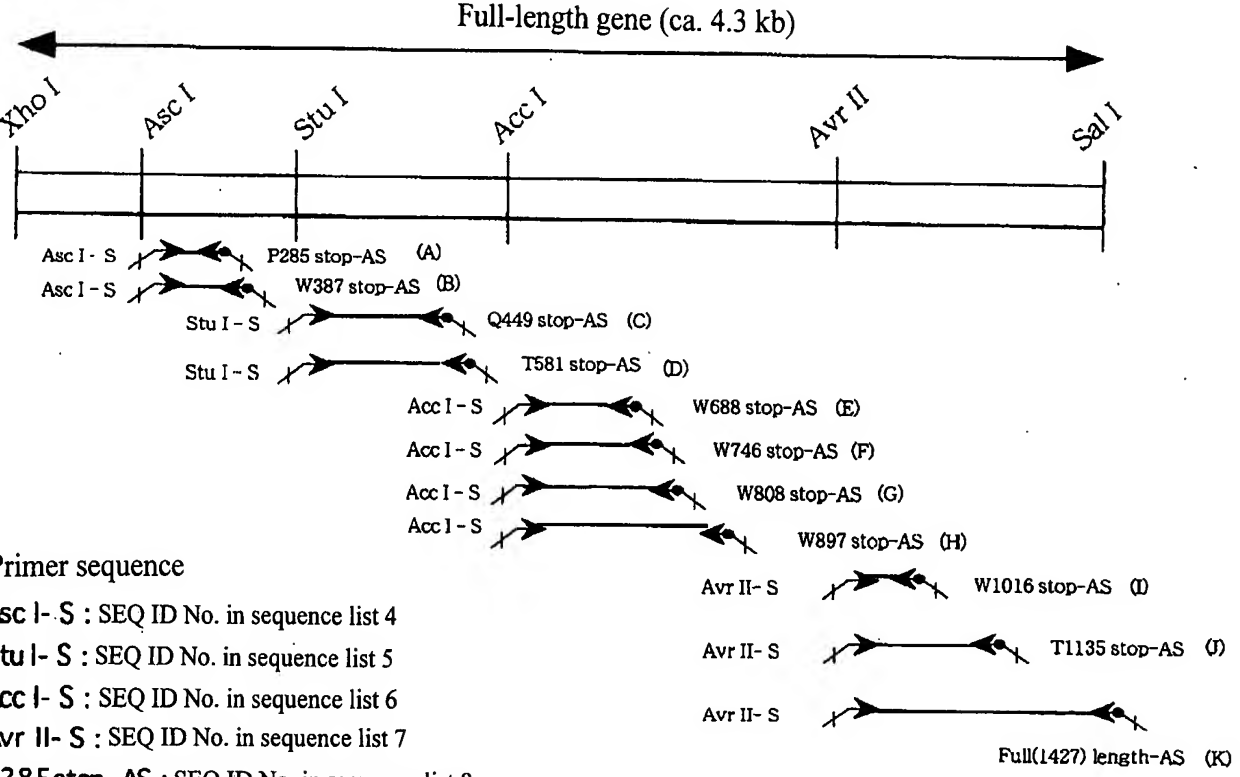
Fig. 1



Primer sequence

Asc I- S : SEQ ID No. in sequence list 4
Stu I- S : SEQ ID No. in sequence list 5
Acc I- S : SEQ ID No. in sequence list 6
Avr II- S : SEQ ID No. in sequence list 7
P285stop- AS : SEQ ID No. in sequence list 8
W387stop- AS : SEQ ID No. in sequence list 9
Q449stop- AS : SEQ ID No. in sequence list 10
T581stop- AS : SEQ ID No. in sequence list 11
W688stop- AS : SEQ ID No. in sequence list 12
W746stop- AS : SEQ ID No. in sequence list 13
W808stop- AS : SEQ ID No. in sequence list 14
W897stop- AS : SEQ ID No. in sequence list 15
W1016stop- AS : SEQ ID No. in sequence list 16
T1135stop- AS : SEQ ID No. in sequence list 17
Full(1427) length- AS : SEQ ID No. in sequence list 18

Antisense primer for preparing C-terminus deleted vector

●: FLAG ; gactacaaggacgatgacgataagtga SEQ ID No. in sequence list 19
AspTyrLysAspAspAspAspLys * SEQ ID No. in sequence list 20

:SalI site

Each of PCR fragments (A) to (K) is ligated.

Fig. 1

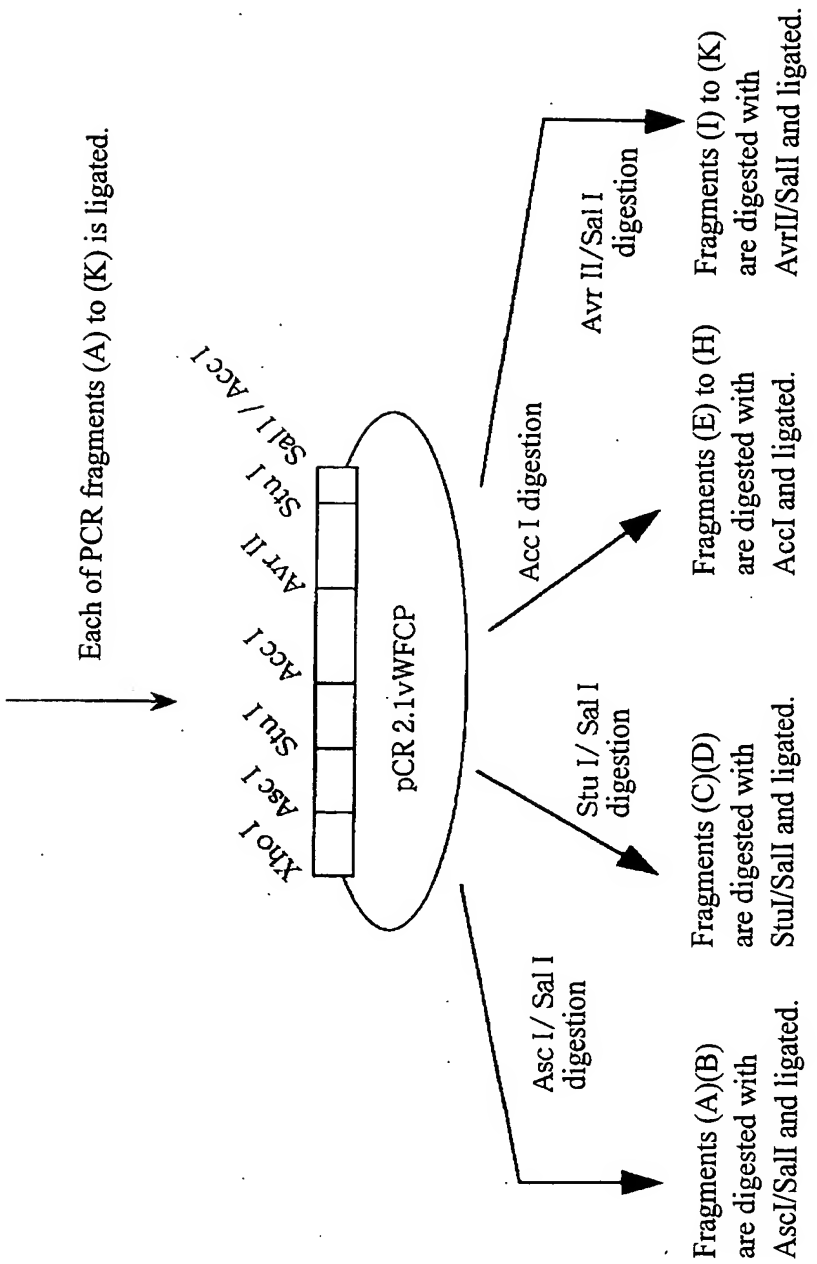
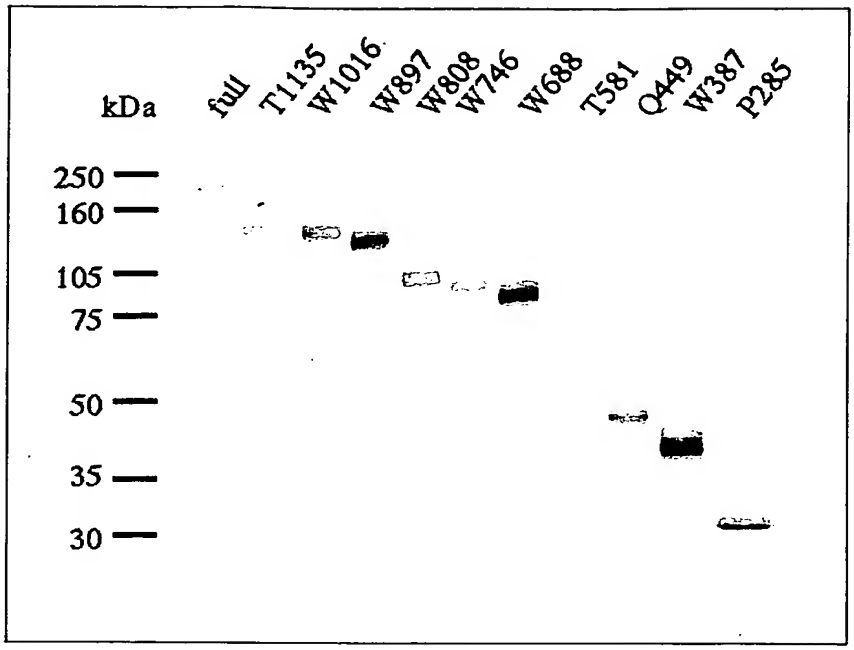

Each of PCR fragments (A) to (K) is ligated.
pCR 2.1vWFCP
Xho I
Asc I
Stu I
Acc I
Avr II
Stu I
Sal I / Acc I
Acc I digestion
Avr II/Sal I digestion
Fragments (I) to (K) are digested with AvrII/SalI and ligated.
Fragments (E) to (H) are digested with AccI and ligated.
Stu I/ Sal I digestion
Fragments (C)(D) are digested with StuI/SalI and ligated.
Asc I/ Sal I digestion
Fragments (A)(B) are digested with AscI/SalI and ligated.

Fig. 2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T1135
W1016
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W808
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W688
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Q449
W387
P285
250
160
105
75
50
35
30

Fig. 3

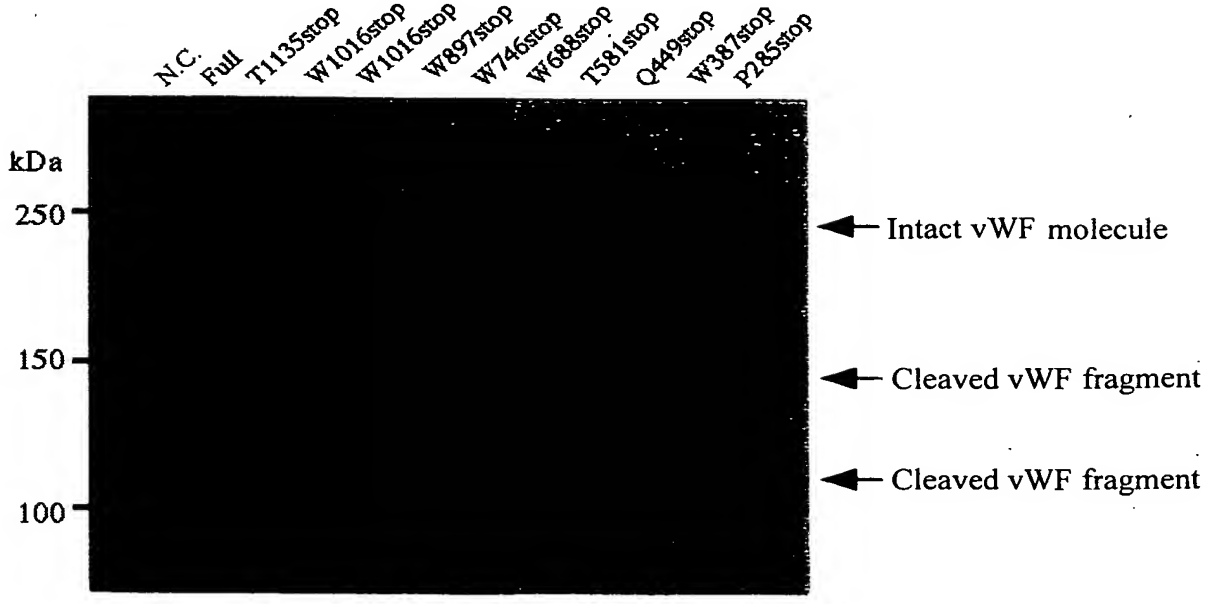
N.C.
Full
T1135stop
W1016stop
W1016stop
W897stop
W746stop
W688stop
T581stop
Q449stop
W387stop
P285stop
kDa
250
150
100
Intact vWF molecule
Cleaved vWF fragment
Cleaved vWF fragment

Fig. 2

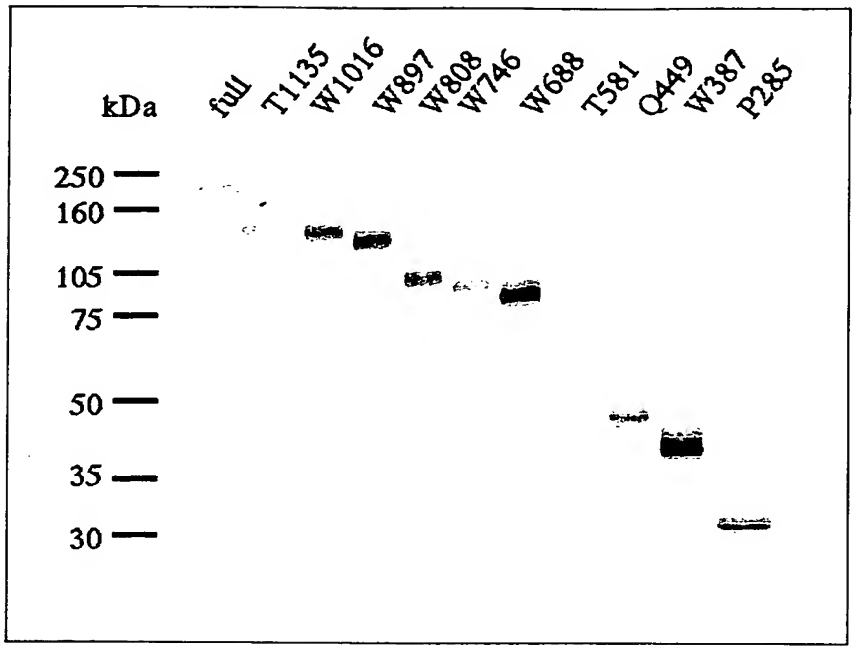

Fig. 3

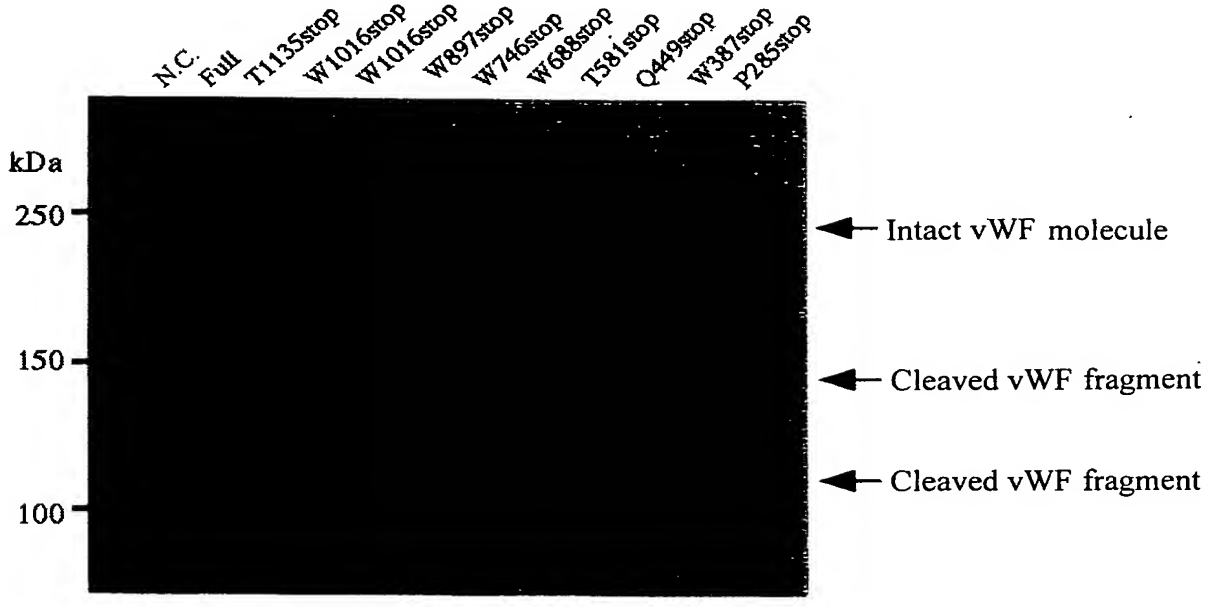

Fig. 2

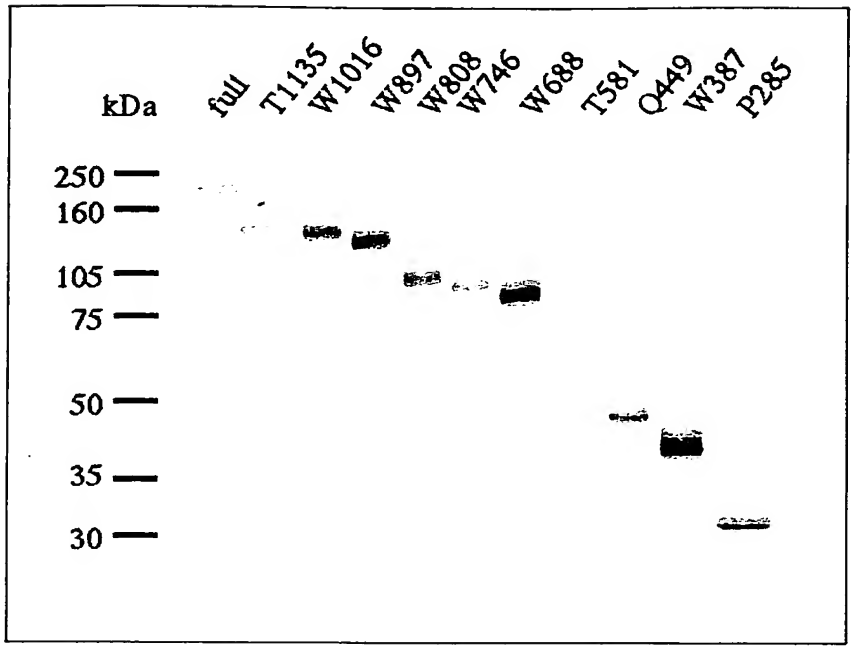
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Fig. 3

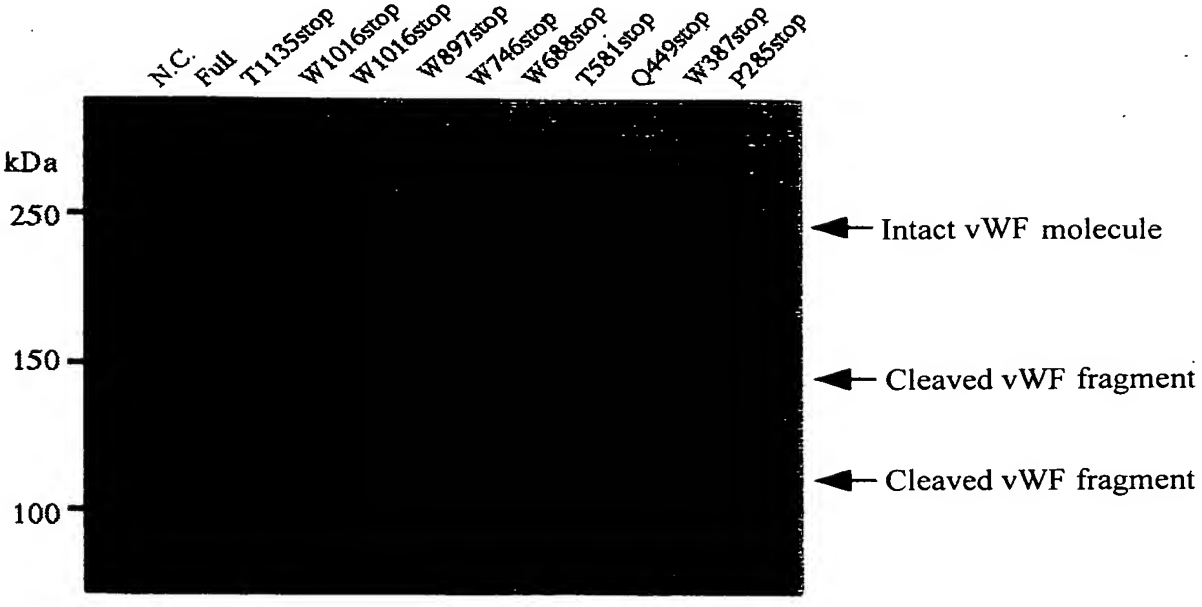
N.C.
Full
T1135stop
W1016stop
W1016stop
W897stop
W746stop
W688stop
T581stop
Q449stop
W387stop
P285stop
kDa
250
150
100
Intact vWF molecule
Cleaved vWF fragment
Cleaved vWF fragment

Fig. 4

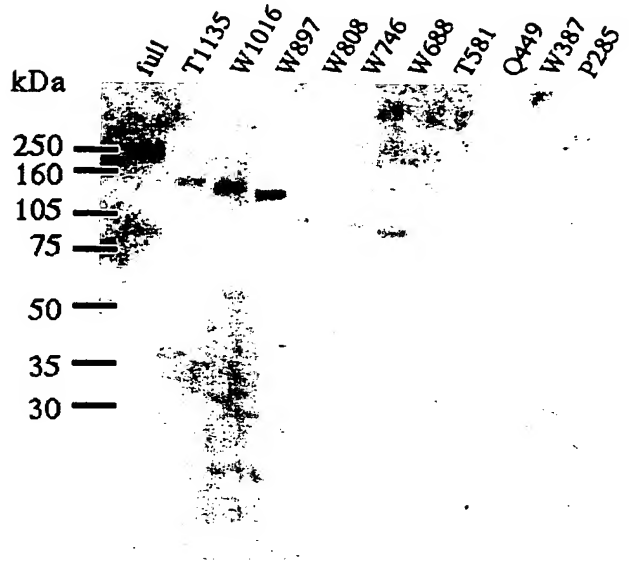
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Fig. 5

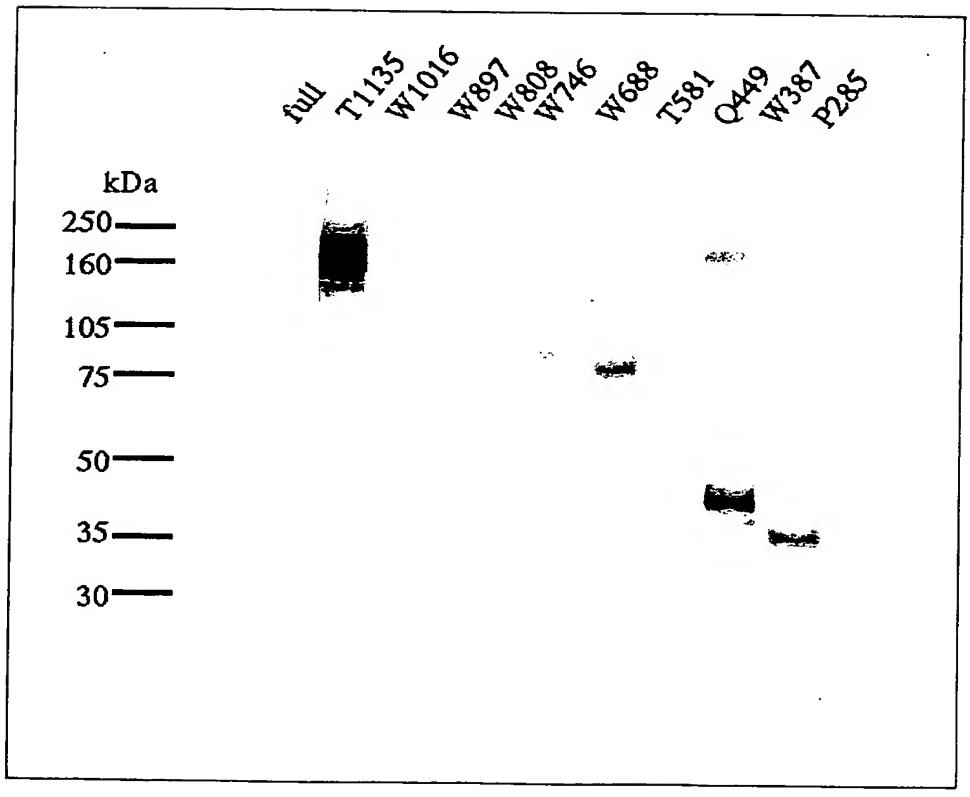
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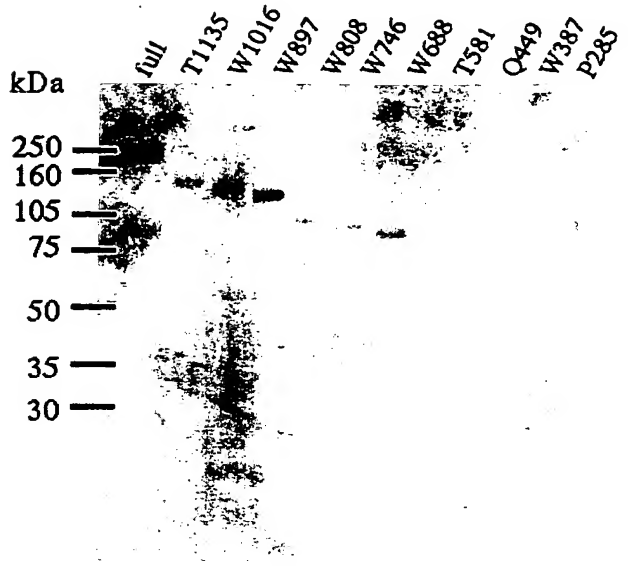
Fig. 4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kDa
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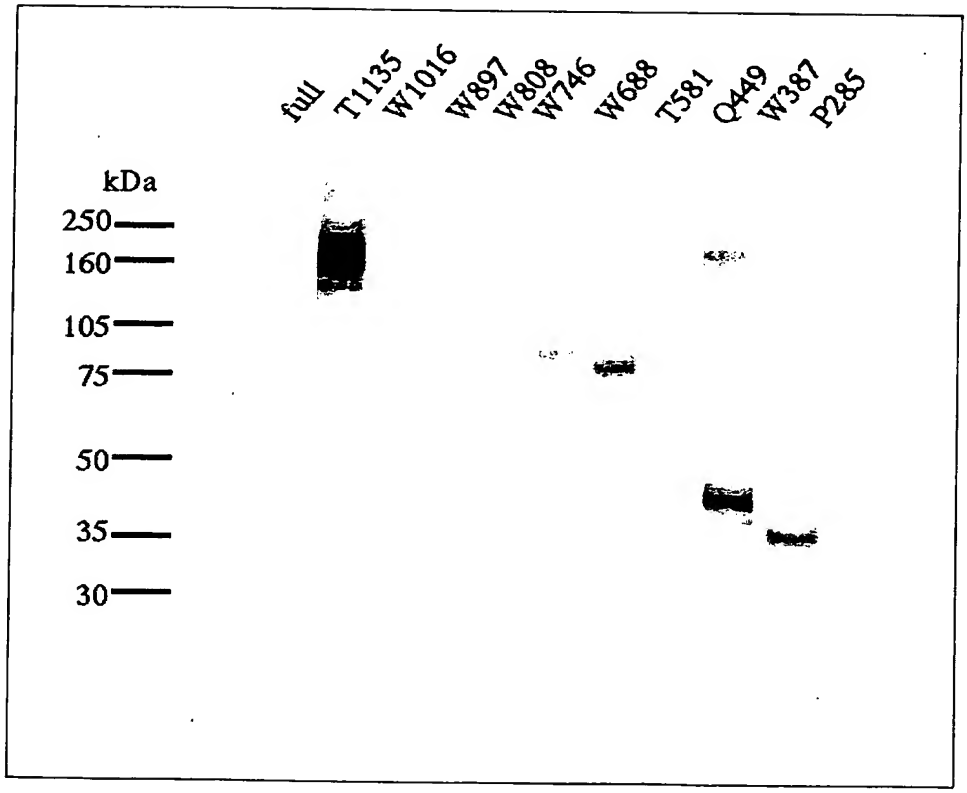
Fig. 5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kDa
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Fig. 4

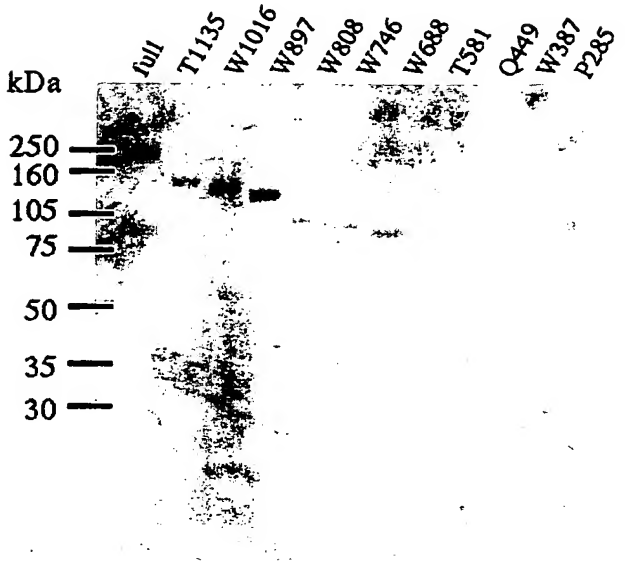
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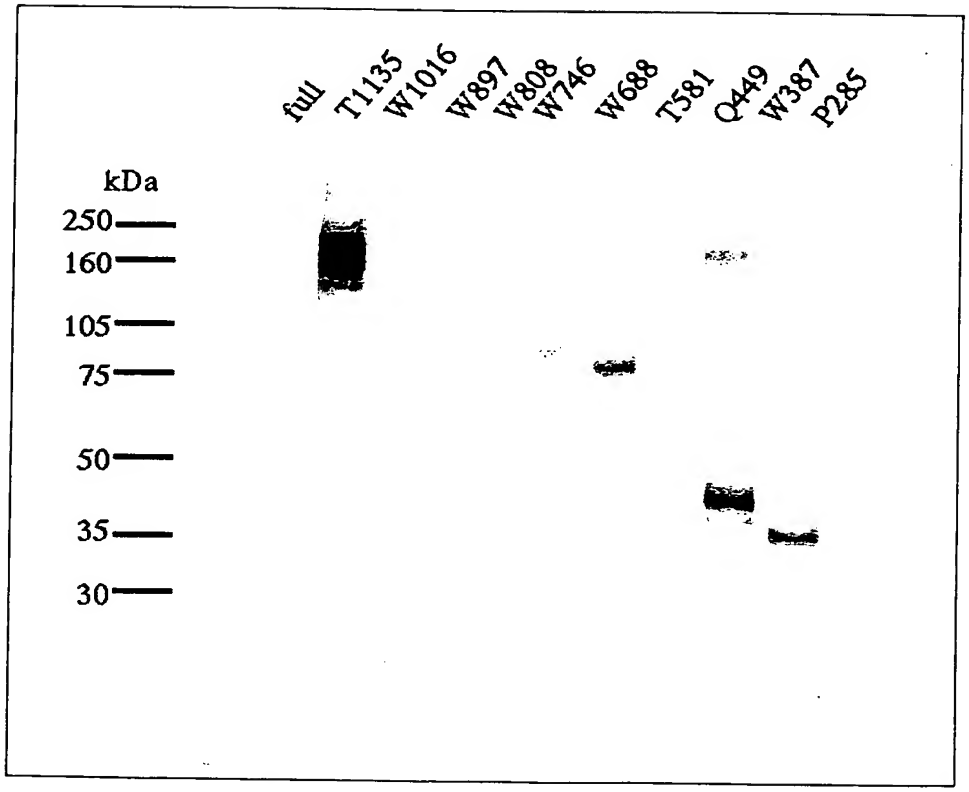
Fig. 5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W1016
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Q449
W387
P285
kDa
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75
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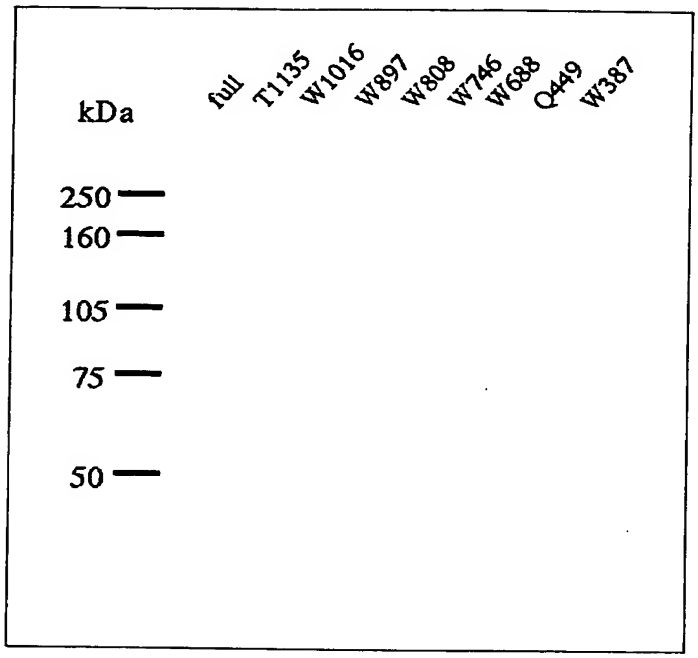
Fig. 6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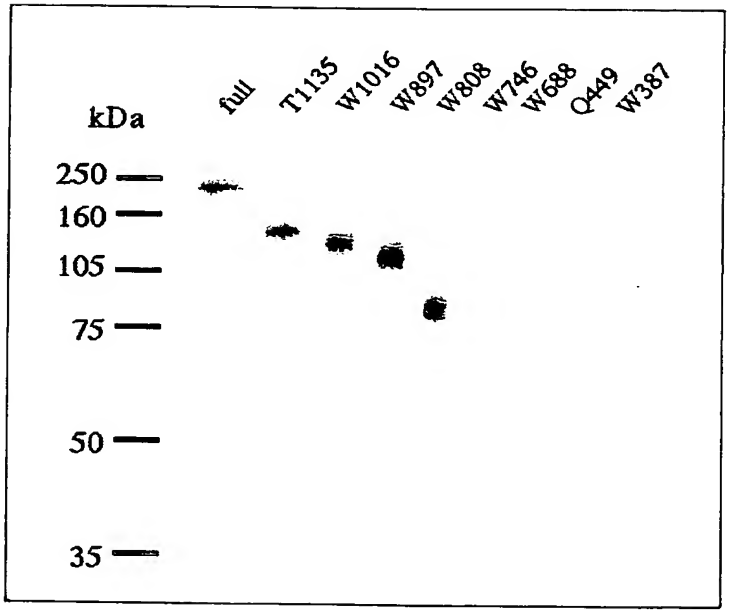
W688
Q449
W387
250
160
105
75
50

Fig. 7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W387
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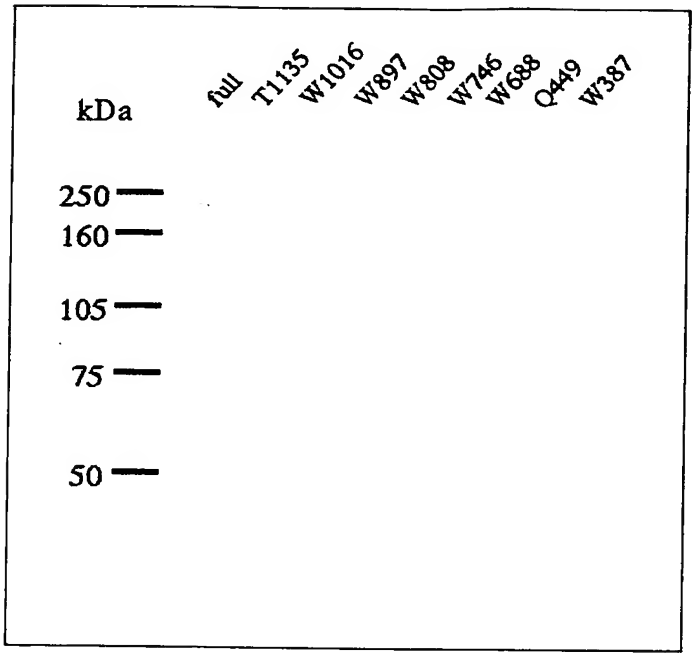
75
50
35

Fig. 6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W1016
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W387
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75
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Fig. 7

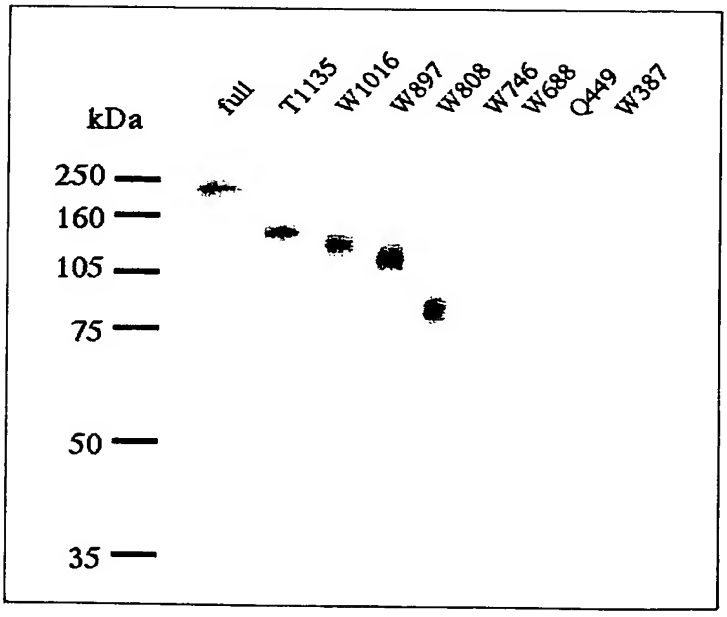
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Fig. 6

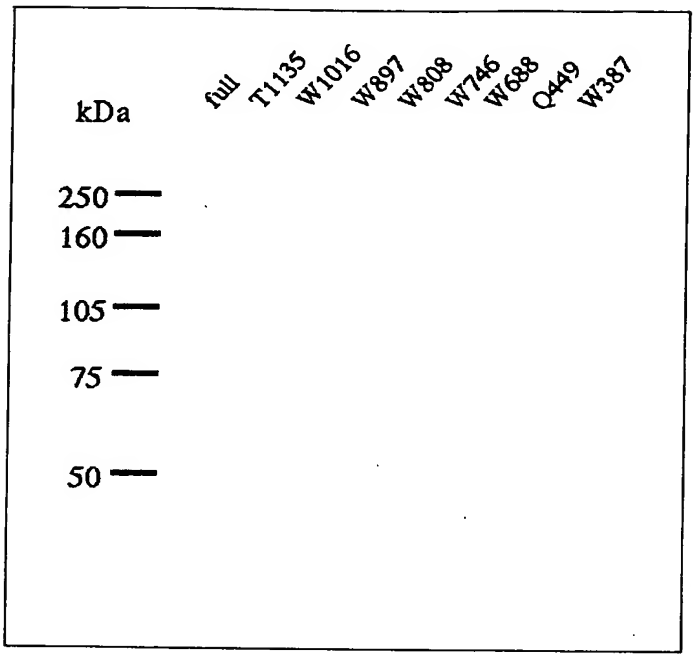
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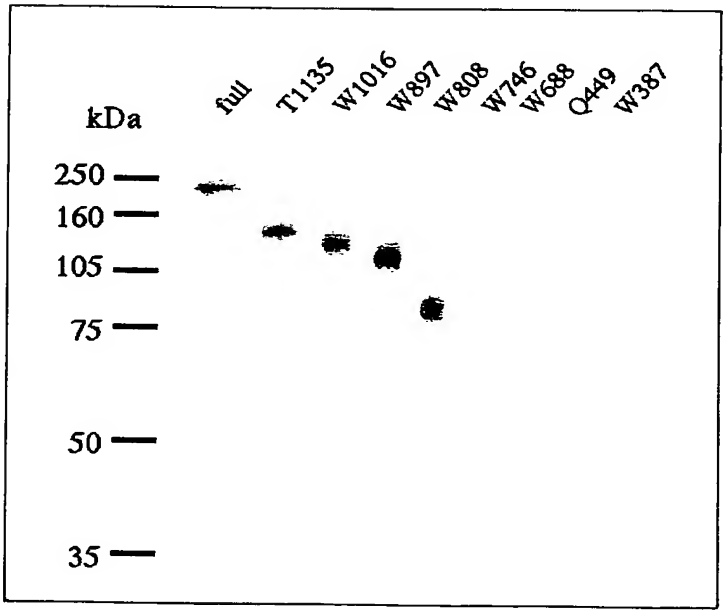
Fig. 7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W808
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W387
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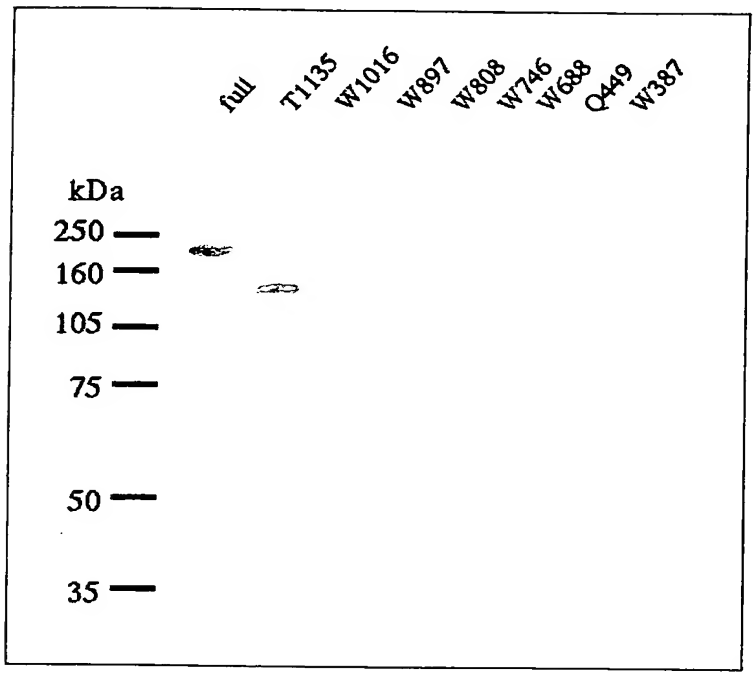
Fig. 8


full
T1135
W1016
W897
W808
W746
W688
Q449
W387
kDa
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160
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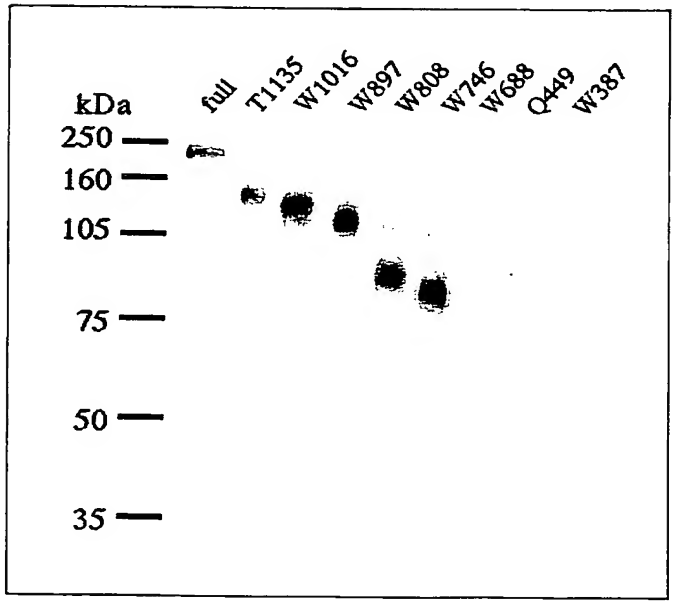
Fig. 9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W897
W808
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Q449
W387
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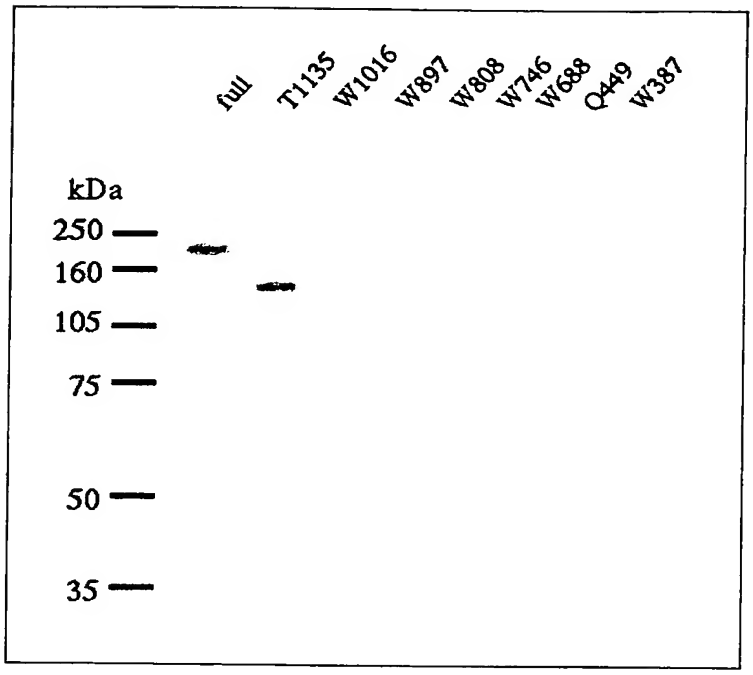
Fig. 8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Q449
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kDa
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Fig. 9

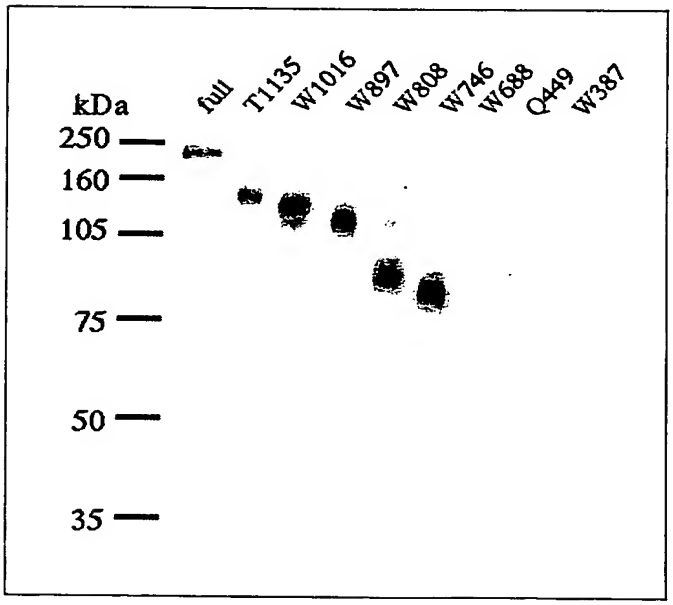
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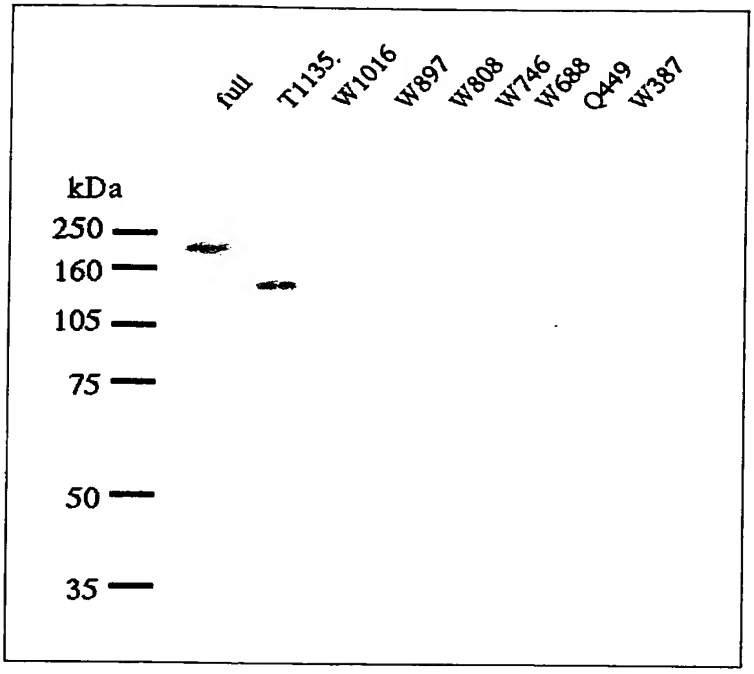
Fig. 8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W808
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Q449
W387
kDa
250
160
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35

Fig. 9

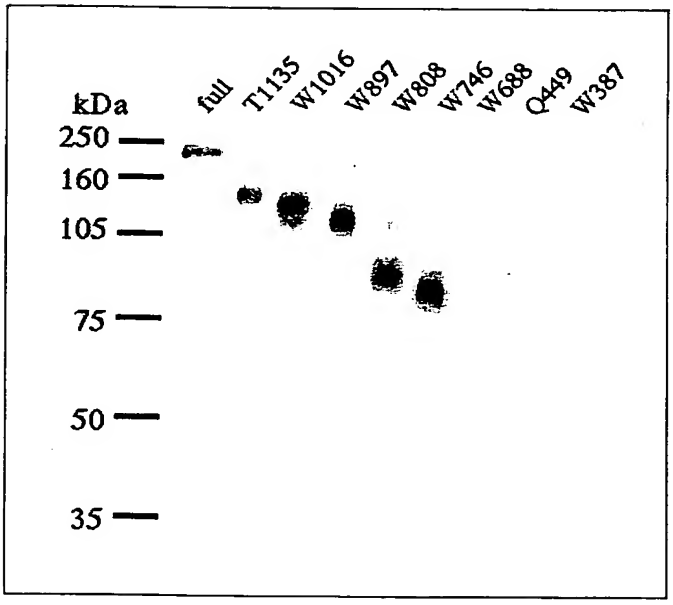

kDa
full
T1135
W1016
W897
W808
W746
W688
Q449
W387
250
160
105
75
50
35

Fig. 10

PoAb 1 Immunization region (Full-length DNA) neutralizing ability +

PoAb 2 Immunization region (Q449stop+FLAG DNA) neutralizing ability +

PoAb 3 Immunization region (P285stop+FLAG DNA) neutralizing ability +

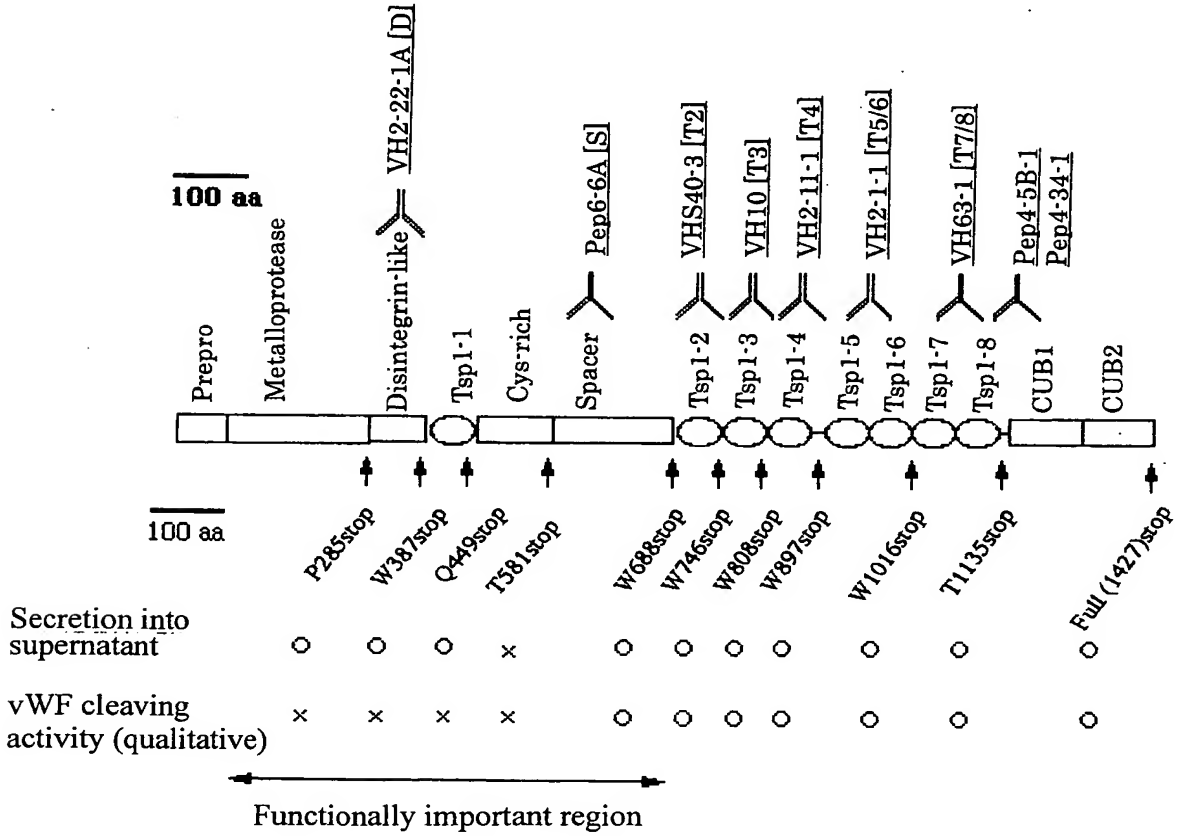
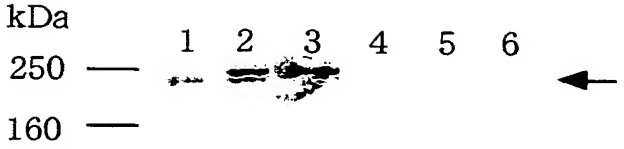
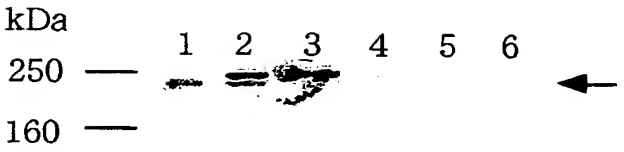


Fig. 11



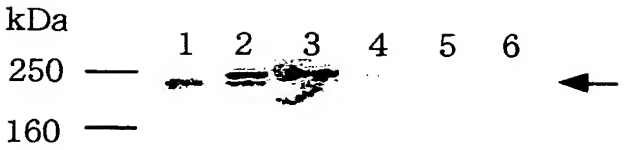
1. Gel filtration sample of FI paste of human pooled plasma
2. Plasma 1 from healthy subject
3. Plasma 2 from healthy subject
4. Plasma 3 from healthy subject
5. Plasma 1 from TTP patient
6. Plasma 2 from TTP patient

Fig. 11



1. Gel filtration sample of FI paste of human pooled plasma
2. Plasma 1 from healthy subject
3. Plasma 2 from healthy subject
4. Plasma 3 from healthy subject
5. Plasma 1 from TTP patient
6. Plasma 2 from TTP patient

Fig. 11



1. Gel filtration sample of FI paste of human pooled plasma
2. Plasma 1 from healthy subject
3. Plasma 2 from healthy subject
4. Plasma 3 from healthy subject
5. Plasma 1 from TTP patient
6. Plasma 2 from TTP patient

Fig. 12

(MoAb-PoAb system)

1. Allow each reagent to be warmed to room temperature
2. Add a sample to WH10 MoAb immobilized plate by 100 μL/well
   37°C, 1 hour
3. Wash the plate with 0.05% Tween-20-TBS three times
4. Dilute PoAb 1 or PoAb 2 with a diluting solution (1% BSA-TBS) so that it may become 1 μg/ml and add to the plate by 100 μL/well
   37°C, 1 hour
5. Wash the plate with 0.05% Tween-20-TBS three times
6. Dilute an anti-rabbit IgG-HRP labelled conjugate with a diluting solution (1% BSA-TBS) to 10000-fold and add to the plate by 100 μL/well
   37°C, 1 hour
7. Wash the plate with 0.05% Tween-20-TBS three times
8. Add a TMB substrate solution (prepared by mixing two liquids at room temperature immediately before use) to the plate by 100 μL/well (positive well turns blue); about 10 minutes (so that the color of 100 ng/ml of the recombinant product enclosed as a standard may be finally about 1 as OD450 nm after the reaction is terminated) at room temperature (positive well turns yellow)
9. Add a reaction terminating liquid (0.5 M sulfuric acid) to the plate by 100 μL/well
10. Measure the plate with a plate reader at 450 nm and 650 nm

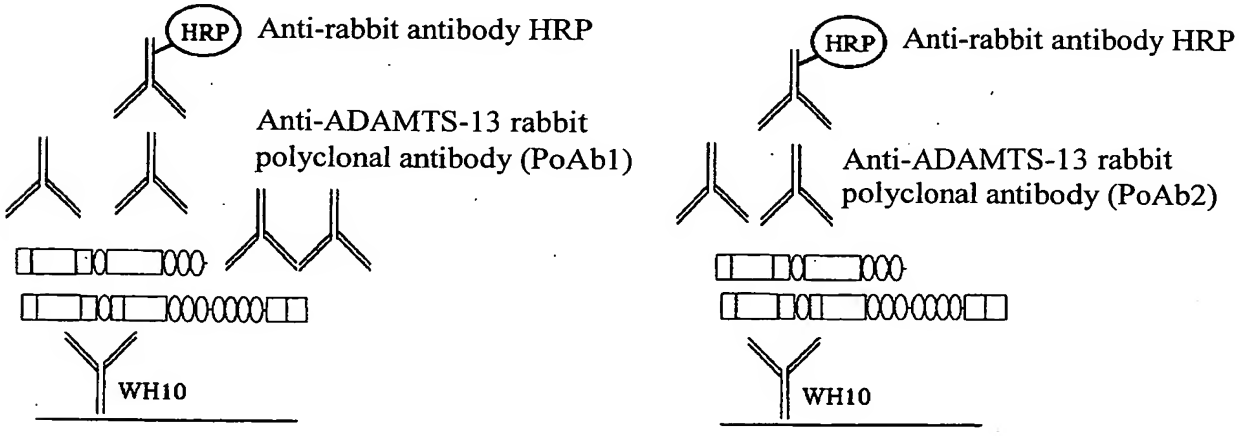
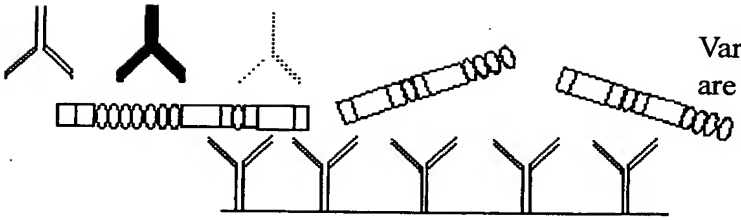


Fig. 13



Various variations of ADAMTS-13 are present in the sample.

Fig. 14

(MoAb-MoAb system)

1. Allow each reagent to be warmed to room temperature
2. Add a sample to WH10 MoAb immobilized plate by 100 μL/well
   37°C, 1 hour
3. Wash the plate with 0.05% Tween-20-TBS three times
4. Dilute a biotinylated antibody with a diluting solution (1% BSA-TBS) so that it may become 1 μg/ml and add to the plate by 100 μL/well
   37°C, 1 hour
5. Wash the plate with 0.05% Tween-20-TBS three times
6. Dilute a streptavidin-HRP labelled conjugate with a diluting solution (1% BSA-TBS) to 10000-fold and add to the plate by 100 μL/well
   37°C, 1 hour
7. Wash the plate with 0.05% Tween-20-TBS three times
8. Add a TMB substrate solution (prepared by mixing two liquids at room temperature immediately before use) to the plate by 100 μL/well (positive well turns blue); about 10 minutes (so that the color of 100 ng/ml of the recombinant product enclosed as a standard may be finally about 1 as OD450 nm after the reaction is terminated) at room temperature (positive well turns yellow)
9. Add a reaction terminating liquid (0.5 M sulfuric acid) to the plate by 100 μL/well
10. Measure the plate with a plate reader at 450 nm and 650 nm.

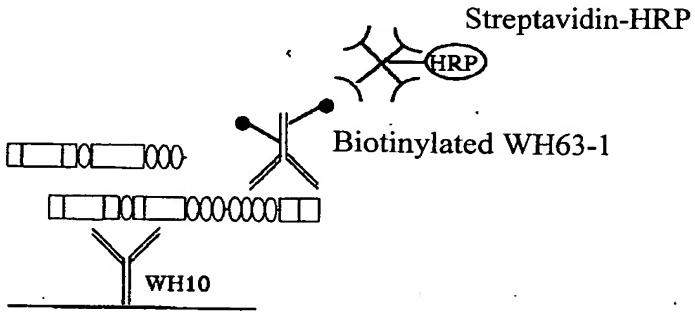


Fig. 15

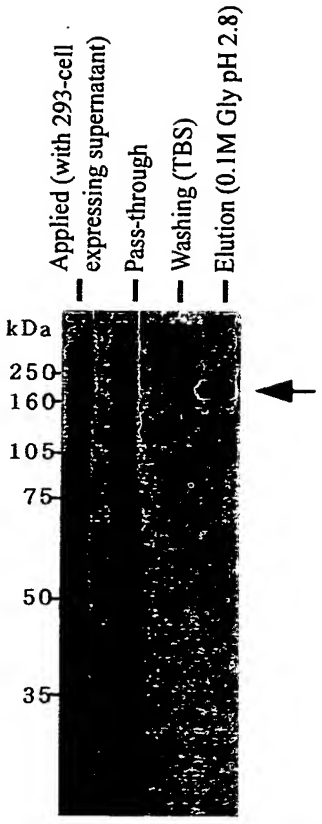

Applied (with 293-cell expressing supernatant)
Pass-through
Washing (TBS)
Elution (0.1M Gly pH 2.8)
kDa
250
160
105
75
50
35

Fig. 15

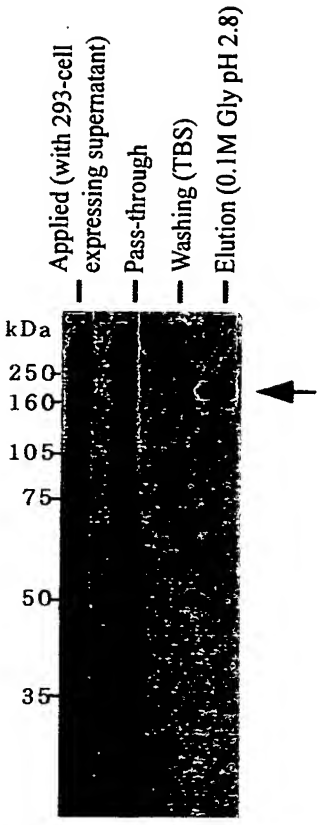

Applied (with 293-cell expressing supernatant)
Pass-through
Washing (TBS)
Elution (0.1M Gly pH 2.8)
kDa
250
160
105
75
50
35

Fig. 15

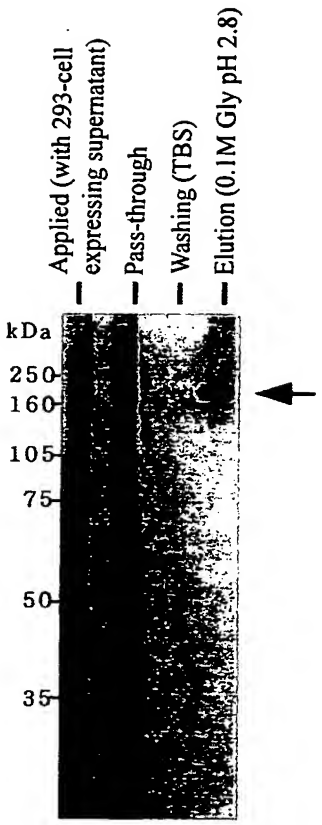

Applied (with 293-cell expressing supernatant)
Pass-through
Washing (TBS)
Elution (0.1M Gly pH 2.8)
kDa
250
160
105
75
50
35

Fig. 16

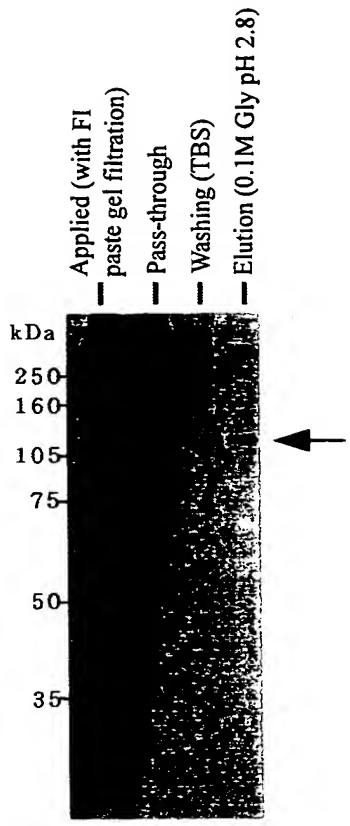

Applied (with FI paste gel filtration)
Pass-through
Washing (TBS)
Elution (0.1M Gly pH 2.8)
kDa
250
160
105
75
50
35

Fig. 16

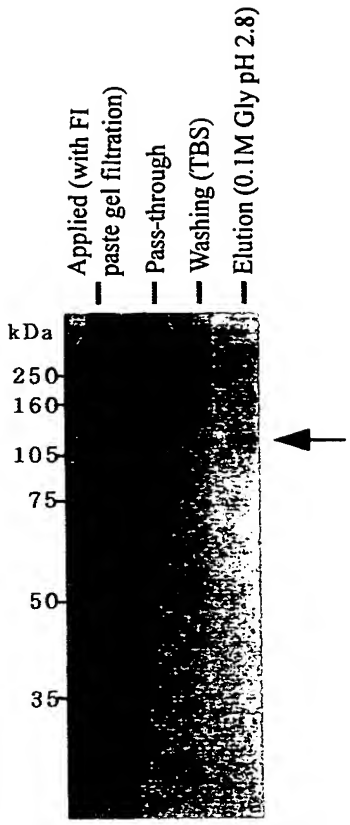

Applied (with FI paste gel filtration)
Pass-through
Washing (TBS)
Elution (0.1M Gly pH 2.8)
kDa
250
160
105
75
50
35

Fig. 16

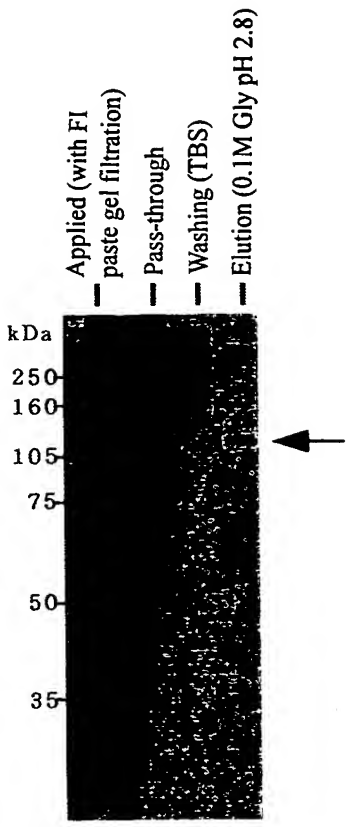
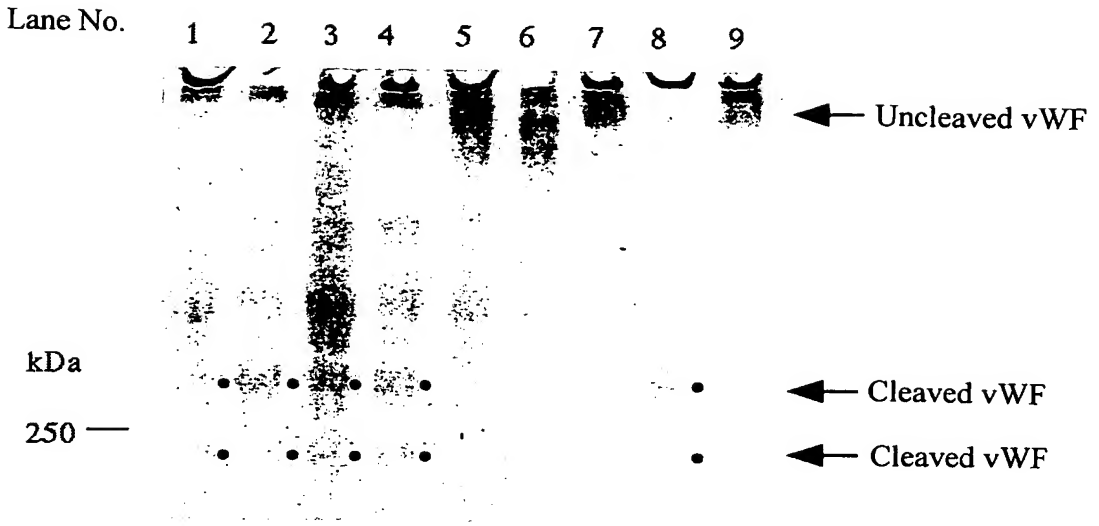
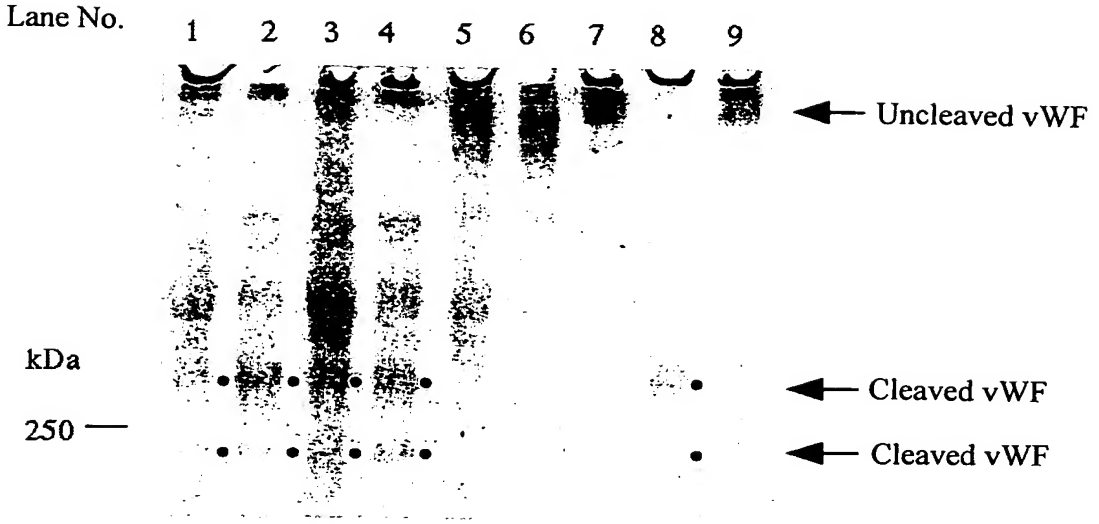

Applied (with FI paste gel filtration)
Pass-through
Washing (TBS)
Elution (0.1M Gly pH 2.8)
kDa
250
160
105
75
50
35

Fig. 17



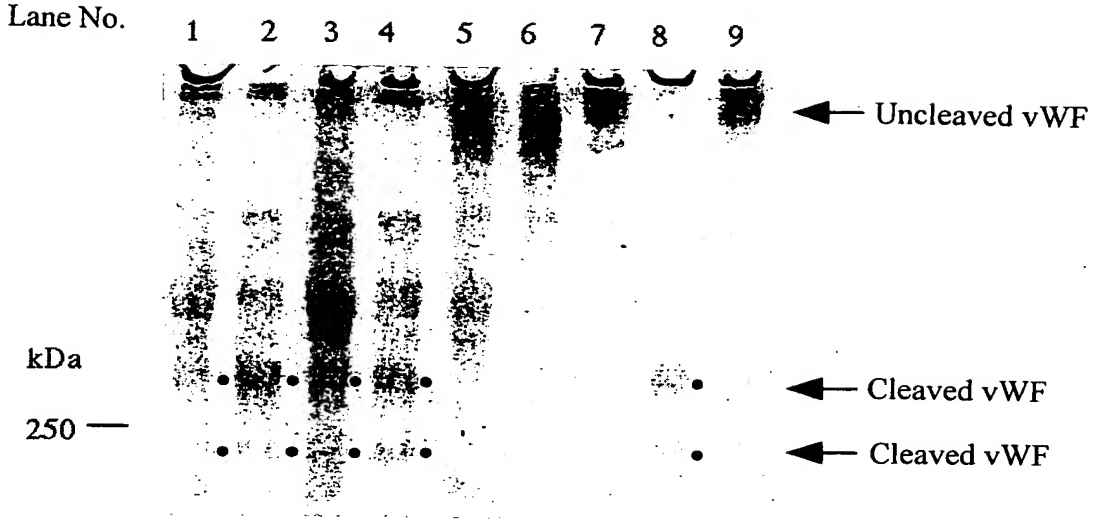
1. ADAMTS-13 solution: normal rabbit serum = 1:1
2. ADAMTS-13 solution: normal rabbit serum (diluted 5-fold)= 1:1
3. ADAMTS-13 solution: peptide immunized rabbit serum = 1:1
4. ADAMTS-13 solution: peptide immunized rabbit serum (diluted 5-fold)= 1:1
5. ADAMTS-13 solution: recombinant protein immunized rabbit serum = 1:1
6. ADAMTS-13 solution: recombinant protein immunized rabbit serum (diluted 5-fold)= 1:1
7. ADAMTS-13 solution: 10 mM EDTA = 1:1
8. ADAMTS-13 solution: buffer only = 1:1
9. Buffer (without ADAMTS-13): buffer = 1:1

Fig. 17



1. ADAMTS-13 solution: normal rabbit serum = 1:1
2. ADAMTS-13 solution: normal rabbit serum (diluted 5-fold)= 1:1
3. ADAMTS-13 solution: peptide immunized rabbit serum = 1:1
4. ADAMTS-13 solution: peptide immunized rabbit serum (diluted 5-fold)= 1:1
5. ADAMTS-13 solution: recombinant protein immunized rabbit serum = 1:1
6. ADAMTS-13 solution: recombinant protein immunized rabbit serum (diluted 5-fold)= 1:1
7. ADAMTS-13 solution: 10 mM EDTA = 1:1
8. ADAMTS-13 solution: buffer only = 1:1
9. Buffer (without ADAMTS-13): buffer = 1:1

Fig. 17



1. ADAMTS-13 solution: normal rabbit serum = 1:1
2. ADAMTS-13 solution: normal rabbit serum (diluted 5-fold)= 1:1
3. ADAMTS-13 solution: peptide immunized rabbit serum = 1:1
4. ADAMTS-13 solution: peptide immunized rabbit serum (diluted 5-fold)= 1:1
5. ADAMTS-13 solution: recombinant protein immunized rabbit serum = 1:1
6. ADAMTS-13 solution: recombinant protein immunized rabbit serum (diluted 5-fold)= 1:1
7. ADAMTS-13 solution: 10 mM EDTA = 1:1
8. ADAMTS-13 solution: buffer only = 1:1
9. Buffer (without ADAMTS-13): buffer = 1:1

Fig. 18

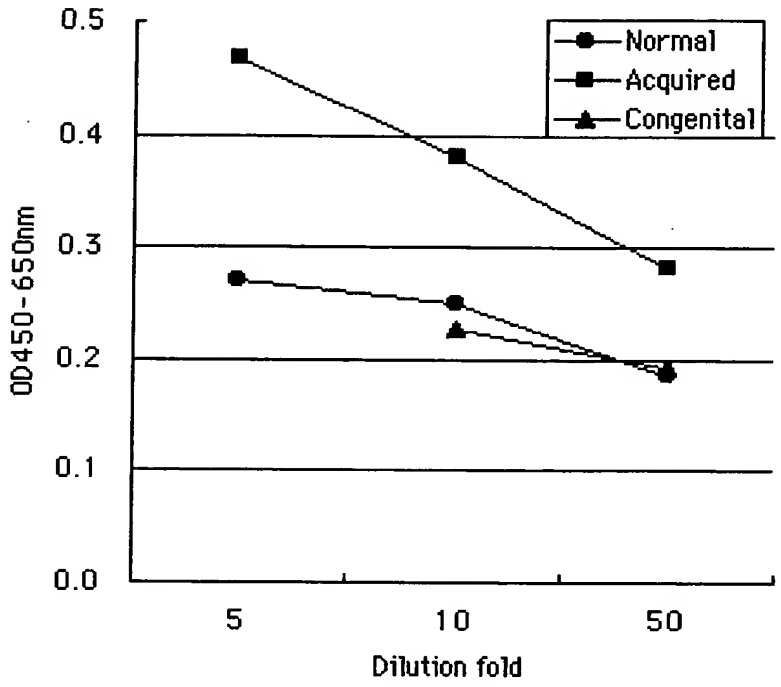

0.5
0.4
0.3
0.2
0.1
0.0
OD450-650nm
Normal
Acquired
Congenital
5
10
50
Dilution fold

Fig. 19

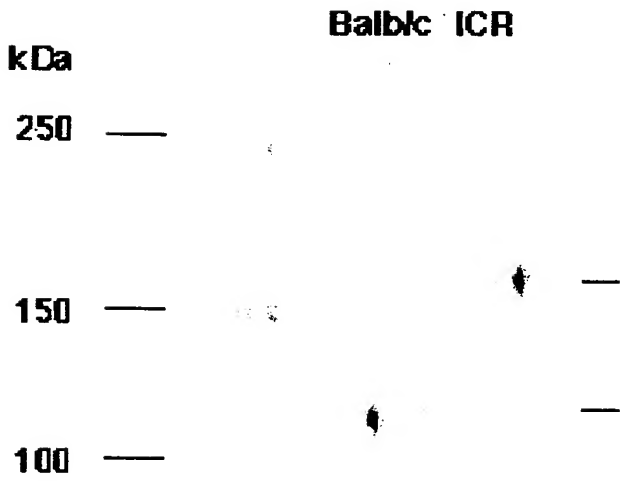


Fig. 19

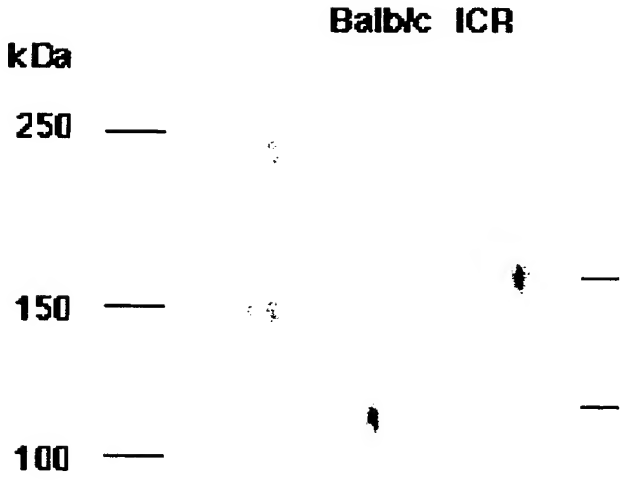


Fig. 19